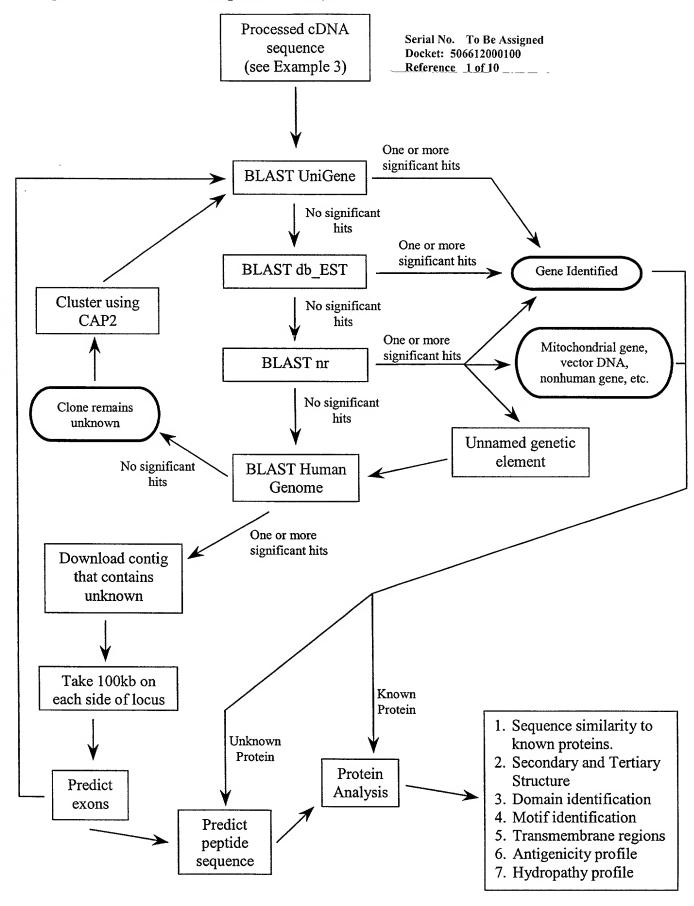
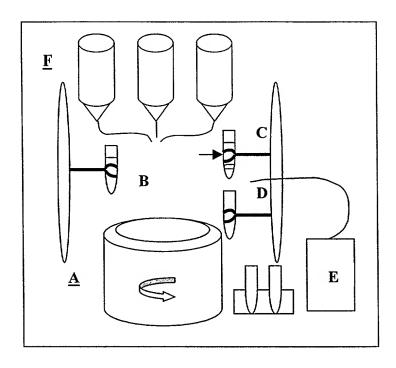
Figure 1: Novel Gene Sequence Analysis



Serial No. To Be Assigned Docket: 506612000100 Reference 2 of 10

Figure 2. Automated Mononuclear Cell RNA Isolation Device



Serial No. To Be Assigned Docket: 506612000100
Reference 3 of 10

Figure 3: Kits for discovery of, or application of diagnostic gene sets

A. Contents of kit for discovery of diagnostic gene sets

- 1. Sterile, endotoxin and RNAse free blood collection tubes (>10cc capacity)
- 2. Alcohol swabs, tourniquet, 18g needle and syringe (>10cc capacity)
- 3. Erythrocyte lysis buffer
- 4. Leukocyte lysis buffer
- 5. Substrates for labeling of RNA (may vary for various expression profiling techniques)

For fluorescence cDNA microarray expression profiling:

Reverse transcriptase and 10x RT buffer

Poly-dT primer

DTT

Deoxynucleotides 100mM each

RNAse inhibitor

Cy3 and Cy5 labeled deoxynucleotides

- 6. cDNA microarrays containing candidate gene libraries
- 7. Cover slips for slides
- 8. hybridization chambers
- 9. Software package for identification of diagnostic gene set from data

Contains statistical methods.

Allows alteration in desired sensitivity and specificity of gene set.

Software facilitates access to and data analysis by centrally located database server.

- 10. Password and account number to access central database server.
- 11. Kit User Manual

B. Contents of kit for application of diagnostic gene sets

- 1. Sterile, endotoxin and RNAse free blood collection tubes (>10cc capacity)
- 2. Alcohol swabs, tourniquet, 18g needle and syringe (>10cc capacity)
- 3. Erythrocyte lysis buffer
- 4. Leukocyte lysis buffer
- 5. Substrates for labeling of RNA (may vary for various expression profiling techniques)

For fluorescence cDNA microarray expression profiling:

Reverse transcriptase and 10x RT buffer

Poly-dT primer

DTT

Deoxynucleotides 100mM each

RNAse inhibitor

Cy3 and Cy5 labeled deoxynucleotides

- 6. cDNA microarrays containing diagnostic gene sets
- 7. cover slips for slides
- 8. hybridization chambers
- 9. Software package for identification of diagnostic gene set from data

Contains statistical methods.

Allows alteration in desired sensitivity and specificity of gene set.

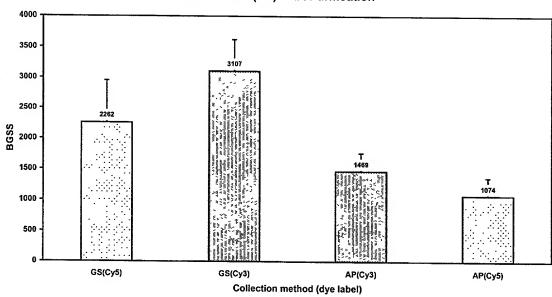
Software facilitates access to and data analysis by centrally located database server

- 10. Password and account number to access central database server.
- 11. Kit User Manual

Figure 4

Serial No. To Be Assigned Docket: 506612000100
Reference 4 of 10

Comparison of Guanine-Silica (GS) to Acid-Phenol (AP) RNA Purification



Serial No. To Be Assigned Docket: 506612000100

Reference 5 of 10

Expression of Leukocyte Specific Genes

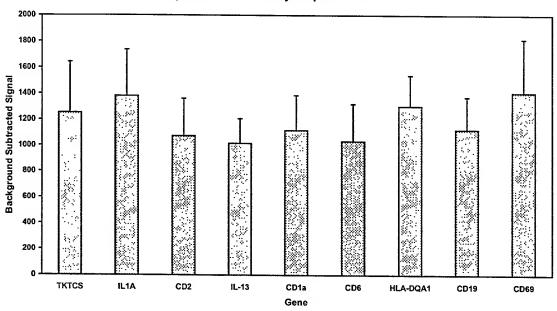
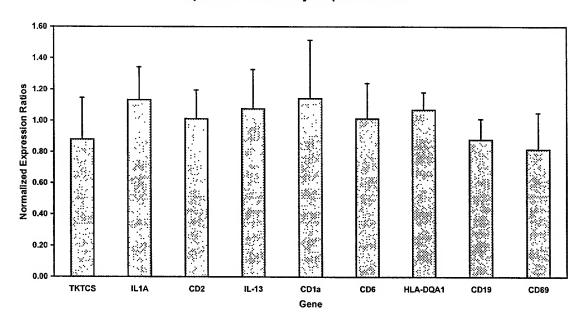


Figure 5

Figure 6

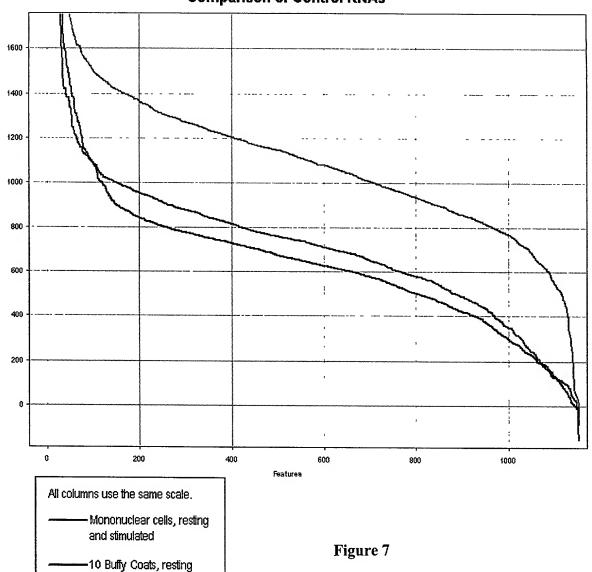
Serial No. To Be Assigned Docket: 506612000100
Reference 6 of 10

Expression of Leukocyte-Specific Genes



Serial No. To Be Assigned Docket: 506612000100
Reference 7 of 10

Comparison of Control RNAs



Mononuclear cells, resting

All markers are connected and ordered by Features.

was labeled.

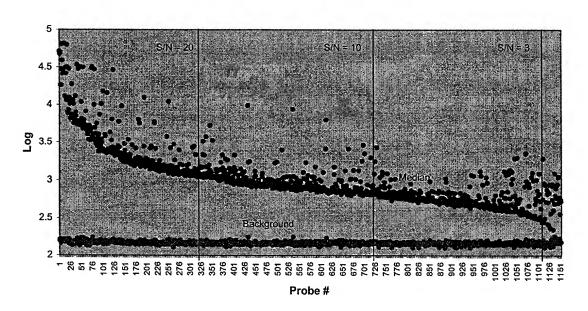
10 µg of each control RNA

Serial No. To Be Assigned Docket: 506612000100

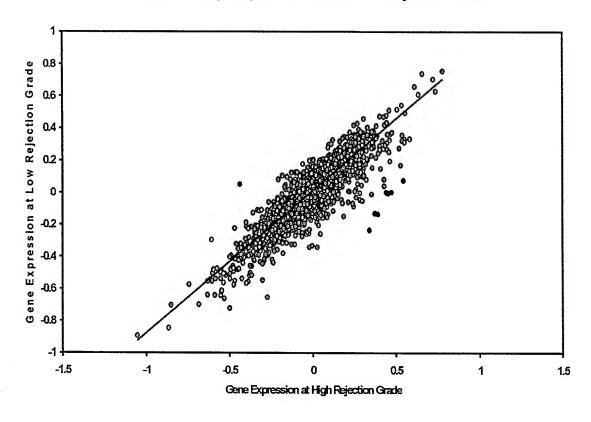
Reference 8 of 10 _____

<u>Figure 8:</u> Log expression of each probe using the R50 reference RNA. Probe expression is ordered by Signal to noise, S/N, decreasing from left to right.

Array Hybe 115018



Comparison of High Rejection Grade to Low Rejection Grade



Serial No. To Be Assigned Docket: 506612000100 Reference 10 of 10

Figure 10: Differential gene expression between grade 0 and 3A samples:

<u>Probe</u>				Array 107742: Grade 0				Array 107739: Grade 3A				Ratio of SRs	
Acc#	Marra.			F532 Median -		SR: scaled	F633 Median -	F532 Median •	Cy3/Cy5	SR: scaled			
NM 003202		Oligo ID	B633	B532	<u>Ratio</u>	ratio (g/r)	B633	B532	Ratio	ratio (g/r)	Grade 0/3A	Grade 3A/0	
BE220959	transcription factor 7 (T-cell specific, HMG-box) (TCF7),	2476			0.188917			358	0.061438	0.219793	3.23048873	0.30955069	
BE220959	major histocompatibility complex, class II, DQ beta 1 (HL	. 6025	1810		0.350829			252	0.117209	0.419312	3.14462275	0.31800317	
NM 002922	major histocompatibility complex, class II, DQ beta 1 (HL		1402					247	0.116455	0.416612	3.13371968	0.31910959	
NM_001781	regulator of G-protein signalling 1 (RGS1), mRNA /cds=	2407	804		0.118159		1884	75	0 039809	0.142415	3.11833431	0.32068403	
NM 002341	CD69 antigen (p60, early T-cell activation antigen) (CD6	2192	4121	405			7385	254	0.034394	0.123043	3.00195843	0.33311587	
BE220959	lymphotoxin beta (TNF superfamily, member 3) (LTB), tr	2283	13488			0.960516	29882	2727	0.091259	0.326476	2.94207495	0.33989617	
NM_001781	major histocompatibility complex, class II, DQ beta 1 (HL	6025	1539			1.257707	1942	237	0.122039	0.436591	2.88074602	0.3471323	
U05040	CD69 antigen (p60, early T-cell activation antigen) (CD6		3850			0.376823		282	0.0366	0.130934	2.87796556	0.34746767	
X14908	far upstream element (FUSE) binding protein 1 (FUBP1	3581	4507	1119		0.933154		220	0.09205	0.329306	2.83369583	0.35289603	
	nuclear receptor subfamily 4, group A, member 2 (NR4A		1365			0.459827	9541	434	0.045488	0.162731	2.82568319	0.35389672	
AF035947	transcription factor 7 (T-cell specific, HMG-box) (TCF7),	2476	2716			0.672539	5310	356	0.067043	0.239845	2.80405488	0.3566264	
	cytokine-inducible inhibitor of signalling type 1b mRNA,	642	9850			2.004771	969	197	0.203302	0.727307	2.75642938	0.36278818	
NM=001781	CD69 antigen (p60, early T-cell activation antigen) (CD6	2192	3357	356	0.106047	0.398574	5963	246	0.041254	0.147586	2.70062225	0.37028503	
					41-		. //						
Y14737	mRNA for immunoglobulin lambda heavy chain /cds=(65	4905	1390	248	0.178417	0.670576	6561	5767	0.878982	3.144527	0.21325167	4.68929496	
Y14737	mRNA for immunoglobulin lambda heavy chain /cds=(65		1398	240	0.171674	0.645231	7159	6112				4.73359863	
BC006402	mRNA for immunoglobulin lambda heavy chain /cds=(65	4481	1826	295	0.161555	0.6072	2973	2498			0.20200364	4.95040579	
X57812	rearranged immunoglobulin tambda light chain mRNA /c	3761	6512	747	0.114711	0.431139		17730	0.647529			5.37301111	
X57812	rearranged immunoglobulin lambda light chain mRNA /c	3761	6728	755	0.112218	0.421766		18636	0.646634		0.18232143		
X72475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3790	8572	1188		0.520889		13892				5.50803866	
X7,2475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3790	15538	2128		0.514739		14245	0.807677			5.61339689	
X72475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3791	11974	1558		0.489034	24261	18761		2.766449		5.65696646	
X57812	rearranged immunoglobulin lambda light chain mRNA /c	3761	6953	778			27621	18560	0.671952			5.71604612	
X72475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3791	10805	1411	0.130588	0.49081	17533			2.924735		5.95900079	
X72475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3790	11246	1453	0.129201	0.4856		13863	0.811936			5 9816215	
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4399	2654	243	0.09156	0.344125		21610	0.57599			5.98789603	
X72475	cDNA: FLJ21321 fis, clone COL02335, highly similar to	3791	10909	1370				18561	0.856609			6.4924922	
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4399	1959	181	0.092394	0.34726		19369	0.63979			6.59109804	
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4399	2558	215	0.08405	0.315899		21936	0.60662		0.14556481	6.86979225	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4474	7538	684		0.341044	6038	4037	0.668599		0.14258368	7.01342553	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4474	8662	780		0.338444		2975	0.685642			7.24745312	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4474	7183	608		0.318133		3909		2.532931	0.12559874	7.96186351	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4475	8986	851		0.355938		1275	0.803403			8.0748531	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4476	11118	1023		0.345828	871			2.801184	0.12364126	8.09993947	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4475	7428	730	0.098277	0.36937	1049	890	0.848427		0.12345771		
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4476		933		0.336757	625	486		2.781837	0.12169477	8.21727973 8.2606647	
BC002963	rearranged immunoglobulin mRNA for mu heavy chain e	4475		484		0.311436		1344	0.793388		0.12105565		
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4398		645		0.304549	22985	18694		2.030319	0.109/2555	9.11364747	
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4398		992		0.304349		12597	0.888991			9.55378803	
AF067420	SNC73 protein (SNC73) mRNA, complete cds /cds=(39	4398			0.072553						0.0980291	10.2010527 11.4716196	